LONG GENERATION TIMES IN BREEDING TREES - A PEST MANAGEMENT BLESSING IN DISGUISE

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ABSTRACT

Although often regarded as a difficulty to be overcome, long generation times in tree improvement may have unrecognized advantages. Long gerneration times have allowed for extensive provenance plantings in which the trees are subjected to a wide variety of pests and diseases under many environmental conditions. A highly touted advantage of the application of biotechnology in tree improvement is a great reduction in this generation time. Forest geneticists may be tempted to abandon extensive field trials prior to release of biotechnologically derived trees. If inadequately tested, such trees may 1) be attacked by new races of pathogens, 2) be highly susceptible to previously unimportant pests or diseases, 3) have lost general resistance to pathogens or pests in the process of selecting specific resistance. 4) have been selected for an in vitro artifact, not disease or pest resistance at all. Forest geneticists should continue use of the time-tested system of extensive provenance plantings and insist that biotechnologically derived materials be so tested prior to wide scale use.

INTRODUCTION

Long generation times may be an unrecognized blessing in tree improvement. In the past, the long generation times needed in tree breeding have been regarded as a difficulty to be overcome. This long generation time in tree breeding has been instead a blessing in disguise, allowing time for extensive field trials and provenance plantings which subject the trees to a wide variety of pests and diseases under many environmental conditions (Heybroek, Stephan and Weissenberg 1982, Van Buijtenen 1984).

This review will emphasize aspects of disease resistance but many of the ideas discussed are equally valid for resistance to pests as well. The reader is referred to Hanover (1982) for a review of insect resistance in trees and to Radwan (1975) for a review of resistance to mammals. The principles which govern development of host resistance are common to both plant and animal hosts and to all types of parasites and predators; factors which cause instability or promote stability in host-parasite systems have many common points (Harlan 1976).

Robinson (1976) provided a highly readable general discussion of the concepts and implications of specific (vertical) and generalized (horizontal) resistance to plant disease and discussed different approaches to disease management. Goodman, Kiraly and Wood (1986,

p.347-415) provide a comprehensive review of mechanisms of disease resistance. They define and describe three main categories of resistance, 1) morphological--which may be on the tissue or organ level, the cellular level or the subcellular level; 2) preformed physiological; and 3) postinfection physiological. Within each type many different host parasite reactions are detailed. Mechanisms of non-specific resistance have also been reviewed (Mullick 1977).

Selection for resistance in plants to biotic agents differs in a fundamental way from selection for resistance or tolerance to abiotic factors. If a plant becomes salt tolerant or cold hardy, that environmental factor does not change in response to the plant's developed resistance. Pests and parasites, on the other hand, can and often do change in response to the host plant changes establishing a genetic feedback relationship; such relationships are believed to exist whenever living organisms interact (Sydor 1976). Introducing specific resistance to a particular pest or pathogen places tremendous selection pressure or that species to evolve to overcome the resistance. One way to prevent the pest or pathogen from overcoming host resistance is to reduce selection pressure by use of partial resistance or tolerance which keeps the problem within acceptable limits but allows a small stable population of the pathogen or pest to remain. This approach is already widely used it pest management strategies for pesticide use; it should provide guidance in selection of host resistance, especially in trees where long term stability of resistance is vital.

In natural forests with native pathogens, epidemics of disease do occur but are not disastrous. Epidemics of disease in natural forests are limited not just by genetic diversity but by 'functional diversity' including discontinuous distributions in time and space, disease tolerance, and both generalized and specific resistance (Schmidt 1978). Robinson (1980) proposed a pathosystem concept for wild plants and its implication for breeding strategies. Deployment of an exotic gene for specific resistance (transplanted from an alien source, for example) into a system which was limited in nature by generalized resistance might set up disastrous consequences.

Orton (1984) reviewed potentials of somaclonal variation and tissue culture methods and explored the underlying principles behind the pheno menon. Wenzel (1985) and Daub (1986) have recently reviewed uses of biotechnology in resistance breeding. Renfroe and Berlyn (1983) described the genetic variation that arises during tissue culture of trees. In addition to use of tissue or cell culture as sources of variation, in vitro methods have been used to grow rust fungi on calli of host trees (Amerson, Frampton and Mott 1985). These dual cultures have been used successfully for screening in vitro for resistance to fusiform rust (Frampton, Amerson and Gray 1985).

Universal adoption of *in vitro* methods, however, would limit deployment of resistance to only those genes detectable in such tests, much reducing the genetic resource available for use. Attempts to use *in vitro* methods to obtain resistance to non-biotrophic tree parasites

have also been tested--one such report is presented in this proceedings (Ostry and Skilling 1987). Tolerance to environmental factors has also been obtained by in vitro selection (Conner and Meredith 1985).

One of the highly touted advantages to the application of biotechnology in tree improvement is the great reduction in generation time--from decades to years or even months. Forest geneticists may be tempted by prospects or pressured by administrators to abandon extensive field trials prior to release of biotechnologically derived trees because, according to Wenzel (1985) "the effectiveness of a new strategy can be measured in terms of release of new varieties" and the new unconventional strategies are expensive.

The chance of resistance succeeding will depend on the mechanism of that response. The various ways in which resistance can occur has been extensively reviewed (Goodman, Kiraly and Wood 1986; Horsfall and Cowling 1980) and will not be so treated here, except to point out that mistaking (or not knowing) the mechanism of resistance could lead to false methodologies in screening in vitro (category #4, below). For example, only a few host-parasite systems have toxins been demonstrated to be the mechanism of pathogenesis (Goodman, Kiraly and Wood 1986), yet this is one of the most often proposed screening mechanisms for in vitro selection of resistance to non-biotrophic pathogens. Daub cautions against attempts to use toxins just because the method is easy (Daub 1986).

In vitro or seedling tests cannot necessarily be relied upon to predict field response in older plants, sometimes such tests are highly predictive, in other cases not (Frampton, Amerson and Gray 1985; Stack 1981, 1985). According to Schmidtling and Walkinshaw (1985) seedling resistance screening for fusiform rust was not highly predictive of field reaction of older plants. Such tests can reduce the size of populations to be field tested but cannot substitute.

Inadequately tested trees may 1) be attacked by new races of pathogens, 2) be highly susceptible to previously unimportant pests or diseases, 3) have lost general resistance to pathogens or pests in the process of selecting specific resistance, 4) may have been selected for an in vitro artifact, not disease or pest resistance at all. These possible ways a new cultivar or line can fail after introduction not hypothetical; all have been documented. For convenience in this discussion I will give each of these cases a name, either taken from an example or describing the situation: 1) The Single Gene Resistance Trap, 2) The Victoria Principle, 3) The Vertifolia Effect, and 4) Missing the Target Completely.

THE SINGLE GENE RESISTANCE TRAP

Single gene resistance (or specific resistance) is most often immunity to prevailing races of the pathogen. Since it allows no disease at all, it looks spectacular beside lines lacking the resistance gene.

Often equally important is that single gene resistance is easy to breed into new varieties and easy to select for in progeny of crosses. These appealing qualities cover a serious flaw. Single gene resistance often lacks durability (Robinson 1976). The wheat breeders were caught by this trap 60 years ago and are still caught. Massive programs by federal and state agencies have been needed for all that time just to keep up with the wheat rust's ability to vary and overcome single gene resistance.

Pathogenic variability appears to be the rule among tree rusts also. Variation in virulence has been demonstrated in poplar rust (Prakash and Heather 1986c), white pine blister rust (McDonald et al. 1984; Kinloch and Byler 1981), fusiform rust (Griggs, Dinus and Snow 1984; Schmidtling 1985) and probably exist in western gall rust (Hoff 1986). To deal effectively with this potential for variability, "An extremely broad genetic base of resistance will be required in each of the host species if tree rusts are to be controlled by the development of resistant strains of" trees (Powers 1982). As early as 1972, concern was expressed for stability of single gene resistance in white pine to blister rust (Hoff and McDonald 1972).

The view expressed by Kinlock and Byler (1981) that "Major gene resistance is highly effective and its deployment.., could be rapidly achieved. Though its durability might be limited by an increase in fre quency of virulent races, we do not consider... the risk great enough to preclude its use, at least for the near term." was made doubtful by the report 3 years later of McDonald et al. (1984) of new races of C. ribi cola after just 13 years of planting trees with single gene resistance.

Because breakdown of single gene resistance is a major problem in agronomic crops, the problem has been extensively studied and documented (Robinson 1976). Using extensive crop data and computer analysis Kiyosawa (1982) concluded that breakdown of resistance occurred mainly in years extremely favorable for the pathogen. Frequency of such years was the biggest factor in longevity of resistance. Studies on poplar rust showed that temperature affects race composition. This means that there will be a genotype by environment interaction for rust on poplar clones planted in climatically different locations (Prakash and Heather 1986b). Races of poplar rust were able to adapt to changed temperatures (Prakash and Heather 1986a). This suggests that the potential exists for unforeseen outbreaks if susceptible clones were planted in areas where rust was present but not damaging on native trees.

Rust race composition and virulence patterns may change as trees are planted in different locations. One might conjecture that a virulent race may "follow" a susceptible host and in its new region pose a threat not just to that host but to other species or clones previously not troubled. It has been proposed that such population shifts have occurred in the recent past by way of explaining present fusiform rust 'Hot Spots' (Schmidtling 1985).

The instability of specific resistance is of particular concern in the present biotechnology climate because this type of resistance is the one most likely (although not the only type) to be detectable in vitro or generated by somaclonal variation or genetic engineering methods.

THE VICTORIA PRINCIPLE

Intensive selection for resistance to a single disease or pest can sometimes result in inadvertent loss of resistance to another. I have used the term Victoria Principle here in reference to one well known example. In the 1930's cereal crop breeders (remember they were already caught in the single gene resistance trap) were looking for sources of resistance to rusts. One particularly good one (Victoria) was found in oats and was bred into new lines, which were widely released. By the early 1940's the Victoria-derived lines began to succumb to a previously unknown disease, which came to be known as Victoria Blight. Later study showed that the causal fungus had always been present but most oats had such high levels of general resistance to it that no noticeable disease occurred. In the process of introducing the rust resistance, a gene for extreme susceptibility to this fungus had also been incorporated, but had gone undetected. Losses were in the hundreds of millions of dollars (Coffman, Murphy and Chapman 1961).

An example of the Victoria Principle in trees is that of the cottonwood cultivar 'Siouxland' which was selected for rust resistance but which is highly susceptible to stem cankers, which severely limit its use (Peterson and Stack 1986). Sometimes such a susceptibility only appears under special circumstances. Francis and McCracken (1985) described a cottonwood clone which looked very promising in early tests but was found to be weakened and predisposed to facultative pathogens after 12 years on a certain soil type. In nature most trees are little affected by most diseases or pests. Many diseases are present but only a few are serious at any one location and time. Clones need disease resistance tailored to the location. "The disease screening process... must include field evaluation in high hazard areas." (Schipper 1976).

Ostry and McNabb (1986) tested poplar clones for 4 diseases. The ratings were done in the second through fourth years at three sites. Many clones were found quite disease susceptible. Resistance to one pathogen did not imply resistance to other diseases. While this test certainly indicated promising candidates for further testing, one result from this test also shows the fault of relying too heavily on short term trials. In their test the Populus cv 'Lombardy' rated very well. On the basis of this test 'Lombardy' might be recommended as a disease resistant cultivar. It is well known however that it is highly susceptible to Dothichiza canker after it reaches larger size and is generally not recommended for that reason.

THE VERTIFOLIA EFFECT

Just as intense selection for resistance to one pathogen may lead to inadvertent loss of resistance to another, so can generalized resistance to a disease be lost if the host is protected by specific resistance. This would not be a problem if specific resistance were permanent, but it is not and when it fails the host may sustain greater damage than a

non-resistant plant. This phenomenon of the erosion of general resistance under an umbrella of specific resistance is called the 'Vertifolia Effect' after one of the most famous examples which occurred in potatoes (Robinson 1976. p. 98-100). A number of examples of this phenomenon are known, some of which have caused great human misery.

Studies on the biochemistry of disease resistance have shed light on why general resistance might be lost. According to McLaughlin and Shriner (1980) most defensive processes cost the plant something, many are expensive. As an example, production of defensive resins ('extractives') by conifers involves".., both de novo synthesis and redistribution of reserves which can lead to.. de reased photosynthesis and decreased growth." Given that situation, selection for growth by a breeder in the absence of disease or pest pressure would likely select less resistant types, since they would not be diverting resources to 'unneeded' defenses.

MISSING THE TARGET

When disease or pest resistance selection is carried out in field plantings, provenance trials or disease nurseries, accurate comparisons can usually be made since they are based on the actual pest or disease of interest--provided it (or they) is present at sufficient levels (Heybroek, Stephan and Weissenberg 1982; Ostry adn McNabb 1986, Schipper 1976). When selection is carried out in vitro on cells or calli, however, a different situation obtains. Artifacts of the tissue culture method need to be guarded against (Renfroe and Berlyn 1983). In one example, selection for resistance to the fireblight bacterium was desired and attempted using cell cultures. Supposedly resistant lines turned out to be artifacts of the culture medium (Bauer and Beer 1985).

CONCLUSIONS

In each of the above mentioned situations, adequate testing in field plantings would have detected the problems. How, then, should forest geneticists handle the matter of selection for resistance in trees? First, they must decide that stability of resistance must be a major, if not the overriding factor in their testing and recognize why this should be so (Schmidt 1978, Sydor 1976).

Deployment of resistance in perennial crops has been analyzed (Robinson 1976, p. 117-122) and breeding programs aimed at stability have been proposed for poplars (Thielges and Land 1976) and other species (Heybroek, Stephan and Weissenberg 1982). Overton and Kang (1985) emphasized the need to retain genetic diversity even in short term breeding programs. While they did not address the questions posed by biotechnology, they did carefully analyze and set forth strong arguments for keeping a diverse base in any improvement program, arguments which are equally valid in the biotech framework.

As described by Schmidt (1978), epidemics of disease in natural forests are limited by 'functional diversity' including discontinuous distributions both of species and of genotypes within species, disease tolerance, and both generalize (horizontal) and specific (vertical) resistance. In use of resistance foresters should attempt to capture the stability of the natural order by planting mosaics of species and a diversity of genotypes within species, genotypes which have differing specific resistance in a background of generalized resistance (Robinson 1980).

"Resistance-mechanisms from nature which are stable ought not to be discarded in favor of exotic untried sources" (Hoff and McDonald 1972). While that statement was made before the era of genetic engineering, the concept is still valid. Nicholls (1979) noted the dangers of too widespread use of red pine, a species with little genetic diversity. Diseases previously not important in the native red pine stands had become a great concern. The same principle applies even more strongly to genotypes within a species. Multiline varieties proposed by crop scientists are an attempt to return to the natural state now enjoyed in forest crops. It would be a foolish mistake to follow the agronomic crop breeders down the road of monoculture and genetic uniformity that even they now admit is a poor choice (Marshall and Pryor 1978, 1979).

Forest geneticists and tree breeders should continue on the time-tested system of extensive provenance plantings and insist that biotechnologically derived materials be so tested prior to wide scale use.

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